

CLAIMS

1. The use of an antigen which is a non-toxic double mutant form of pertussis toxin for the manufacture of a vaccine composition for intranasal administration to induce an immune response against *B.pertussis* infection.
2. The use of a non-toxic double mutant form of pertussis toxin for the manufacture of an adjuvant composition for stimulating or enhancing a protective immune response of an antigen co-administered therewith.
3. The use according to Claim 2 wherein the composition is for administration to a mucosal surface.
4. The use according to Claim 3 wherein the composition is for intranasal administration.
5. The use according to any one of the preceding Claims wherein the non-toxic double mutant form of pertussis toxin is one in which the glutamic acid 129 amino acid in the  $S_1$  sub unit has been substituted by another amino acid.
6. The use according to Claim 5 wherein the glutamic acid 129 amino acid has been substituted by glycine.

7. The use according to any one of the preceding Claims wherein the arginine 9 amino acid has been substituted.
8. The use according to Claim 7 wherein the arginine 9 amino acid has been substituted by lysine.
9. The use according to Claim 1 wherein the vaccine composition contains one or more other pertussis antigens selected from filamentous haemagglutinin (FHA) and the P69 outer membrane (P69).
10. The use according to Claim 9 wherein the non-toxic double mutant form of pertussis toxin is as defined in any one of Claims 5 to 8.
11. The use according to Claim 9 or Claim 10 wherein the vaccine composition contains both FHA and P69.
12. The use according to Claim 2 wherein the said antigen is the C-fragment of tetanus toxin.
13. The use according to Claim 2 wherein the non-toxic double mutant form of pertussis toxin is as defined in any one of Claims 5 to 8.
14. A vaccine composition adapted for intranasal administration, the vaccine composition comprising a

non-toxic double mutant form of pertussis toxin, and a pharmaceutically acceptable carrier.

15. A vaccine composition according to Claim 14 wherein the non-toxic double mutant form of pertussis toxin is one in which the glutamic acid 129 amino acid in the S<sub>1</sub> sub unit has been substituted by another amino acid.
16. A vaccine composition according to Claim 15 wherein the glutamic acid 129 amino acid has been substituted by glycine.
17. A vaccine composition according to any one of Claims 14 to 16 wherein the arginine 9 amino acid has been substituted.
18. A vaccine composition according to Claim 17 wherein the arginine 9 amino acid has been substituted by lysine.
19. A vaccine composition according to any one of Claims 14 to 18 which contains one or more other pertussis antigens selected from filamentous haemagglutinin (FHA) and the P69 outer membrane (P69).
20. A vaccine composition according to Claim 19 which contains both FHA and P69.

21. A vaccine composition comprising an antigen and an adjuvant capable of enhancing the immune response to the antigen in a mammal to which the composition is administered; characterised in that the adjuvant is a mutant form of pertussis toxin as defined in any one of Claims 14 to 18.
22. A vaccine composition according to Claim 21 in which the antigen is tetanus toxin C fragment.
23. A vaccine composition according to Claim 21 or Claim 22 which is adapted for administration to a mucosal surface, and in particular the nasal mucosa.
24. A vaccine composition according to any one of Claims 14 to 23 in the form of nasal drops or a nasal spray.
25. A vaccine composition according to any one of Claims 14 to 24 packaged in a container adapted to dispense a metered dose of the composition in spray or drop form.
26. A method of immunising a host such as a mammal (e.g. human) against B.pertussis infection, which method comprises administering to the host intranasally an effective amount of a composition as defined in any one of Claims 14 to 25.

27. A method of stimulating or enhancing an immune response to an antigen in a mammal; which method comprises co-administering with the antigen an effective adjuvant amount of a non-toxic double mutant form of pertussis toxin.
28. A method according to Claim 27 wherein the Glu 129 amino acid in the S<sub>1</sub> sub-unit of the pertussis toxin has been substituted by another amino acid.
29. A method according to Claim 27 wherein the antigen and the non-toxic mutant form of pertussis toxin are administered to a mucosal surface of the mammal.
30. A method according to any one of Claims 27 to 29 wherein the glutamic acid 129 amino acid in the S<sub>1</sub> sub-unit has been substituted by glycine.
31. A method according to any one of Claims 27 to 30 wherein the non-toxic mutant form of pertussis toxin is a double mutant in which the arginine 9 amino acid residue has been substituted by another amino acid.
32. A method according to Claim 31 wherein the arginine 9 amino acid has been substituted by lysine.
33. A method according to any one of Claims 27 to 32 wherein the antigen and the non-toxic form of

pertussis toxin are administered intranasally.

34. A method according to any one of Claims 27 to 32 wherein the antigen and the non-toxic mutant forms of pertussis toxin are administered at the same time.
35. A method according to Claim 34 wherein the antigen and the non-toxic mutant form of pertussis toxin are present in admixture in a composition administered to the mammal.
36. A method according to any one of Claims 27 to 35 wherein the antigen is the C-fragment of tetanus toxin, or one or more immunogenic fragments thereof.

In the Claims

Please cancel without prejudice claims 1-36. Please add the following new claims.

37. A method of stimulating or enhancing a protective immune response to an antigen in a mammal; which method comprises co-administering to a mucosal surface of said mammal with the antigen an effective adjuvant amount of a non-toxic double mutant form of pertussis toxin, said antigen being one which elicits a protective immune response when co-administered with said effective adjuvant amount of non-toxic double mutant form of pertussis toxin.
38. A method according to claim 37 wherein the non-toxic double mutant form of pertussis toxin comprises an S<sub>1</sub> sub unit containing an amino acid at position 129 which is other than glutamic acid.
39. A method according to claim 38, wherein the amino acid at position 129 in the S<sub>1</sub> sub-unit is glycine.
40. A method according to claim 37 wherein the non-toxic double mutant form of pertussis toxin comprises an S<sub>1</sub> sub unit containing an amino acid at position 9 which is other than arginine.
41. A method according to claim 40 wherein the amino acid at position 9 is lysine.
42. A method according to claim 37 wherein the antigen and the non-toxic form of pertussis toxin are administered intranasally.
43. A method according to claim 37 wherein the antigen and the non-toxic double mutant form of pertussis toxin are administered at the same time.
44. A method according to claim 43 wherein the antigen and the non-toxic double mutant form of pertussis toxin are present in admixture in a composition administered to the mammal.
45. A method according to claim 37 wherein the antigen is selected from the group consisting of tetanus toxin C-fragment, and one or more immunogenic fragments thereof.

46. A method according to claim 45 wherein the vaccine composition contains both FHA and P69.

47. A vaccine composition in the form of nasal drops or a nasal spray, the composition comprising an antigen and an adjuvant capable of enhancing the immune response to the antigen in a mammal to which the composition is administered; wherein the adjuvant is a non-toxic double mutant form of pertussis toxin and said antigen is one which elicits a protective immune response when co-administered with said adjuvant.

48. A vaccine composition according to claim 47 wherein the mutant form of pertussis toxin comprises an S<sub>1</sub> sub unit containing an amino acid at position 129 which is other than glutamic acid.

49. A vaccine composition according to claim 48 wherein the amino acid at position 129 is glycine.

50. A vaccine composition according to claim 47 wherein the non-toxic double mutant form of pertussis toxin comprises an S<sub>1</sub> sub unit containing an amino acid at position 9 which is other than arginine.

51. A vaccine composition according to claim 50 wherein the amino acid at position 9 is lysine.

52. A vaccine composition according to claim 47 packaged in a container for dispensing a metered dose of the composition in spray or drop form.

53. A vaccine composition comprising an antigen and an adjuvant which enhances the immune response to the antigen in a mammal to which the composition is administered; wherein the adjuvant is a non-toxic double mutant form of pertussis toxin and the antigen is selected from tetanus toxin C fragment and one or more immunogenic fragments thereof.

54. A vaccine composition according to claim 53 which is in a form selected from nasal drops and a nasal spray.

REMARKS

Please enter this amendment prior to calculation of the fees. Applicant has amended the claims to more distinctly claim the invention. No new matter has been added by any of the foregoing.

Respectfully submitted,



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